Induction of rotational behaviour by intranigral baclofen[1] suggests possible GABA-agonist activity

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Summary. In rats, unilateral injections of the GABA-derivative baclofen into the zona reticulata of the substantia nigra produced a contralateral rotation that was translated to ipsilateral rotation under the influence of amphetamine. These results mimic those following unilateral elevation of GABA levels in the substantia nigra and suggest that baclofen may have some GABA agonist activity following intracerebral injection.

There is considerable debate as to whether baclofen, a structural analogue of γ-aminobutyric acid (GABA), possesses GABA-agonist activity[3,4]. In early iontophoretic studies baclofen [β-(p-chlorphenyl) GABA] was shown to produce a depression of neuronal activity that was, however, insensitive to the GABA antagonist bicuculline[5,6]. More recently, baclofen has been shown to produce bicuculline-sensitive, GABA-like effects on lumbo-sacral motoneurons[7].

Rotational behaviour (circling)[8] can be induced by unilateral elevation of GABA levels in the zona reticulata of the substantia nigra (SNR); rotations are ipsilateral under the influence of amphetamine[9], and contralateral in the absence of amphetamine[10]. These effects are presumed to be mediated via a striatonigral modulation by GABA neurones of activity in dopamine neurones ascending from the nigra to the striatum[11], whose asymetric activation underlays rotation[8]. We have made SNR injections of baclofen and find similar rotational responses.

Materials and methods. Male Sprague-Dawley rats (150 ± 20 g) were anaesthetized with ether and given unilateral stereotaxic[12] injections of baclofen (100 ng in 1 µl saline) or saline (1 µl) into SNR. Rats were then placed in automated rotometer bowls for continuous recording of rotations. For further groups of rats these injections were made following a 45 min pretreatment with d-amphetamine (5 mg/kg) and rotations similarly recorded. Subsequent to testing rats were decapitated, their brains removed, fixed in formol-saline for 1 week and then subjected to conventional histological procedures for verification of injection sites. Statistical comparisons between groups were made using the Student's t-test (2-tailed).

Results. Control injections of saline into SNR induced a weak rotation; similar injections of baclofen induced a moderate contralateral rotation that was significantly different from rotations of control animals ($p < 0.01$) (figure 1). When these injections were made into SNR of amphetamine-pretreated animals saline induced a mild

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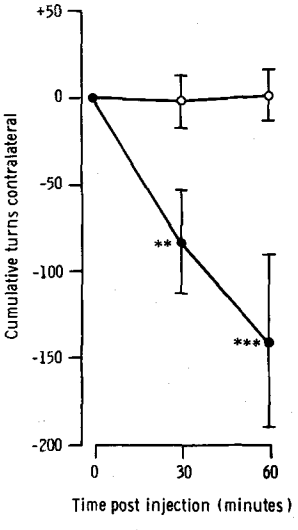

Fig. 1. Rotational responses following unilateral intranigral injections of 100 ng baclofen in 1 µl saline (N = 6) (●) or 1 µl saline (N = 10) (○). Results are expressed as cumulative rotations ± SEM against time following injection. Significant different from control values: ** $p < 0.02$; *** $p < 0.01$.

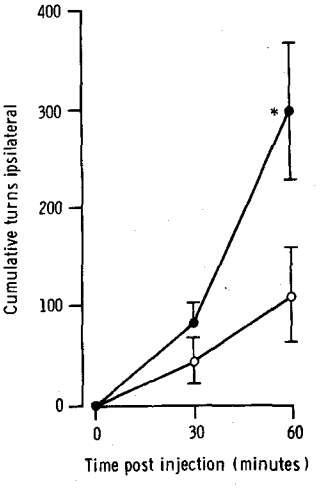

Fig. 2. Rotational responses following unilateral injections into substantia nigra of rats treated 45 min previously with d-amphetamine 5 mg/kg. Animals received either baclofen, 100 ng in 1 µl saline (N = 10) (●) or 1 µl saline (N = 6) (○). Results are expressed as in figure 1. Significant difference from control value: * $p < 0.05$.

ipsilateral rotation; baclofen induced a significantly more intense ipsilateral circling response ($p < 0.05$) (figure 2). Histological examination revealed that injection sites were within SNR.

Discussion. In untreated and amphetamine-pretreated rats unilateral injections of baclofen into SNR produce contralateral and ipsilateral rotation respectively. These rotational responses are very similar to those seen after unilateral elevation of nigral GABA levels in both untreated[10] and amphetamine-pretreated[9] animals. While the nature of the interaction between striatonigral GABA neurones and nigrostriatal dopamine neurones within the substantia nigra is contentious[11], these effects following injections into SNR suggest that baclofen may have some GABA agonist activity following direct intracerebral injection. Though original reports that baclofen may be an antagonist of substance P[13] have failed to be confirmed subsequently[14,15], the recent demonstration of striatonigral pathways to SNR containing substance P[16] as well as GABA suggests that mechanisms for these present baclofen effects in terms of interactions with neurotransmitter systems other than those involving GABA cannot, however, be excluded.

Baclofen has been widely used as a putative GABA-mimetic substance both in animal studies[17] and in the clinic[18], without convincing evidence for GABA agonist activity. Its use in such experiments has, however, provided circumstantial evidence for GABA-ergic properties in that baclofen has been shown to mimic the effects of elevation of brain GABA levels on both animal[19] and clinical[20] models of nigrostriatonigral mechanisms. The wide range of pharmacological properties of baclofen[21,22] necessitates further study aimed at clarifying its status as a hypothetical GABA-mimetic agent.

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The relative sensitivity of pulmonary parenchymal cells to ^{239}plutonium dioxide[1]

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Summary. Alpha particles inhaled by mice affect primarily type II epithelial cells, whereas interstitial mononuclears, alveolar macrophages and type I epithelium are much more resistant and apparently react secondarily. The cellular responses, qualitative and quantitative, exhibit a time-dose relationship.

Materials and methods. A2G mice were exposed for 10 min to inhalation of a $^{239}PuO_2$ aerosol generated by an exploding wire technique in a specially constructed chamber[2]. In separate experiments the animals, in groups of 12, received estimated lung doses of approximately 22 nCi or 150 nCi, respectively referred to as the low and high dose series. Control mice were given a mock exposure in the same apparatus. Animals survived for 3–26 weeks after inhalation. Mice from control and both test groups were killed in pairs by i.p. pentobarbital and lung tissue embedded in araldite for electron microscopy. In addition to analyzing qualitative changes, differential counts of alveolar wall cells were made from electron micrographs, whose number and range of magnification were standardized for all groups. Throughout the period of observation radioactivity, as detected by X-ray measurements and autoradiographs of 1 μm sections, persisted in the lungs.

Results. The type II alveolar epithelial cell is essentially secretory in function[3], a main product being surfactant, which is associated with the osmiophilic lamellar bodies[4]. In normal mice these bodies were commonly vacuolated but their rounded outlines betrayed the former content of secretory material (figure 1,a). The number of type II cells in both test groups rose by 50% and vacuolated bodies became fewer though larger. In high-dose animals the subcellular changes were more severe and from 15 weeks pronounced alterations affected about half the population. The cytoplasm was then distended by large smooth spaces containing a little lamellar material and deforming the nuclei. Over some vacuoles the remaining cytoplasm was extremely thin (figure 1,b) and had ruptured onto the alveolar surface. As a late feature of high-dose mice, some cells developed changes suggesting regeneration, as evidenced by flattening, small and scanty but denser lamellar bodies and by intact mitochondria and endoplasmic reticulum.

Alveolar macrophages normally possess phagosomes (figure 2,a). Low-dose mice showed no quantitative change but the phagosomes tended to be larger. In high-dose mice, especially from the 10-week-interval, the number of macrophages rose and their cytoplasm was crowded with phagosomes or inclusions (figure 2,b), some of which strongly suggested lamellar bodies discharged by type II cells. Degenerative changes were not apparent. Interstitial cells (figure 1,a), distinct from fibroblasts, may be regarded as emigrated monocytes residing temporarily in the pulmonary interstitium before emerging onto the alveolar surface as mature macrophages. No qualitative changes were detected but,

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